IJPSR (2025), Volume 16, Issue 11 (Research Article)

INTERNATIONAL JOURNAL OF PHARMACEUTICAL SCIENCES AND RESEARCH

Received on 09 April 2025; received in revised form, 07 June 2025; accepted, 14 June 2025; published 01 November 2025

# FORMULATION AND DEVELOPMENT OF RIMEGEPANT LOADED *IN-SITU* NASAL GEL FOR TREATMENT OF MIGRANE BY EXTENDED-RELEASE DRUG DELIVERY

Hemalata Sagar Wadkar *, Krutika Sanjay Nilwani and Shital Purbha Narwade

Department of Pharmaceutical Quality Assurance, Abhinav Education Society's College of Pharmacy, Narhe, Pune - 411041, Maharashtra, India.

**Keywords:**

Rimegepant, Extended-release drug delivery, *In-situ* nasal gel, Poloxamer 407, Gellan Gum, Migraine treatment

**Correspondence to Author:**
**Dr. Hemalata Sagar Wadkar**

Associate Professor,
Department of Pharmaceutical Quality Assurance, Abhinav Education Society's College of Pharmacy, Narhe, Pune - 411041, Maharashtra, India.

**E-mail:** hemabhargude@yahoo.com

**ABSTRACT:** Migraine treatment efficacy is often limited by conventional oral delivery systems. The objective of this study was to formulate and optimize a thermoreversible in-situ nasal gel of Rimegepant to provide extended drug release and improve therapeutic effectiveness. Rimegepant-loaded thermoreversible in-situ nasal gels were formulated using varying concentrations of Poloxamer 407 and Gellan Gum through factorial design. Formulations were characterized for physicochemical properties, thermal behavior (DSC), drug-excipient compatibility (FTIR), gelation characteristics, mucoadhesive strength, and *ex-vivo* permeation using goat nasal mucosa. The optimized formulation (KF8) exhibited ideal characteristics with gelation temperature of 35.23±0.45°C, gelation time of 9.76±1.4 seconds, viscosity of 51.76±0.96 m.Pa. s, and mucoadhesive strength of 6972.2±19.39 dyne/cm². DSC analysis confirmed drug purity (melting point 175.53°C) and compatibility with excipients. Ex- vivo studies demonstrated sustained drug release with 98.31±4.92% permeation over 10 hours, flux of 12.51 µg/cm²/h, and permeability coefficient of 0.00167 cm/h. The developed thermoreversible in-situ nasal gel demonstrates promising potential for efficient Rimegepant delivery, combining optimal gelation characteristics, sustained release, and warranting further clinical investigation for migraine treatment.

## INTRODUCTION:

***In-situ* Nasal Gel:** Gel transformation from liquid form occurs naturally at targeted treatment areas through specific body signals using the advanced technology of *in-situ* gelation [1]. A gel forms out of solution through interactions with components like temperature shifts, pH speedups, or ion concentration based on both formulation design and gelation mode.

QUICK RESPONSE CODE

DOI: 10.13040/IJPSR.0975-8232.16(11).2954-68

This article can be accessed online on www.ijpsr.com

DOI link: https://doi.org/10.13040/IJPSR.0975-8232.16(11).2954-68

The adaptive properties of *in-situ* gels create better benefits compared to basic gel products [2]. Prefabricated gels prove challenging to distribute evenly and need supplementary equipment while *in-situ* gives you fluid first with simple application. The treatment method becomes easier with negligible product waste while delivering uniform protection across the nasal cavity. After application the formulation reacts with its environment to swell and hold a stable position across the mucosal surface which enhances drug delivery techniques [3].

**Mechanisms of *In-situ*:** Gel Formation In-situ gelation is the ultimate technique that enables the separate liquid phases of formulations to solidify into a gel phase in response to specific physiological stimuli.

This mechanism relies on knowingly the dynamic characteristics of polymers, that allows the formulation to be applied as a liquid so that the substance can spread evenly at the targeted area and then solidify into a gel form which fixate on the mucosal lining [4].

The gel formation process offers a possibility of increased drug loading, retention time, and management of drug release and therapeutic advantages. The primary methods of *in-situ* gelation involve temperature-sensitive gelation, pH-sensitive gelation, and ion sensitive gelation with each method employing a different condition [5].

**Temperature-Sensitive Gelation:** Some polymers are responsive to temperature; at 37°C they change their sol state to gel like state. These polymers become liquid at lower temperatures, say at room temperature but, exhibit a gel like properties at or above the critical solution temperature or the gelation temperature. Three most commonly used polymers are Poloxamer 407 (Pluronic F127). Solution temperature or gelation temperature [6]. Poloxamer 407 (Pluronic F127), A widely used thermosensitive polymer. It is a triblock copolymer containing polyethylene oxide (PEO) segment and polypropylene oxide (PPO) segment. At low temperatures, the preferential interaction of the hydrophilic PEO blocks is responsible for 3 keeping the polymer in a liquid state. At elevated temperature, the PPO blocks become hydrophobic and cause micelle formation and gelation of the gelatos [7].

**pH-Sensitive Gelation:** pH-sensitive gelation relies on polymers that remain in a liquid state at certain acidic or basic pH values but form a gel when the pH changes to physiological levels. The ionization or protonation of functional groups within the polymer structure results in the formation of a three-dimensional network, resulting in gelation. Carbopol, A cross-linked polyacrylic acid that gels at near-neutral pH [8]. In acidic environments, it remains in a compact, coiled form. Upon exposure to the nasal cavity's neutral pH (approximately 6.8–7.0), it ionizes, expanding and forming a gel. Chitosan, A natural polymer derived from chitin. It remains soluble in acidic conditions but forms a gel when neutralized [9].

**Ion-Sensitive Gelation:** Ion-sensitive gelation is based on the polymers which are sensitive to certain ions in the physiological medium. These polymers swell and/or swell and engage ionic cross-linking once they come into contact with divalent or multivalent cations such as calcium or magnesium. This mechanism also favorable for nasal delivery since such ions are inherent in mucus of the nasal cavity [10]. Gellan gum, A hydrocolloidal, heterogeneous non- gelling polysaccharide that gels in the presence of divalent cations. Its gelation results from ionic cross-linking of calcium ions with carboxylate groups in the polymer chain. Other ion- sensitive polymer is sodium Alginate which gels in the presence of calcium ions. The formation of covalent and ionizable bonds between the alginate chains and calcium result in a sturdy, mucoadhesive gel network [11].

**Polymers used in *In-situ* Nasal Gel:** Polymers are the building blocks of *in-situ* nasal gel formulations and are responsible for the sol to gel transition property under physiological conditions. These materials have critical roles to fold in terms of the formulation's stability, gelation, and mucoadhesiveness besides controlling drug release. Polymers used in *in-situ* nasal gels can be broadly categorized based on their gelation triggers: These include temperature sensitive, pH sensitive and ion sensitive applications. This section outlines the most frequently used polymers in these systems according to their classification based on the chemical structure [12].

Poloxamer 407 (Pluronic F127) is the most commonly studied thermosensitive polymer in in situ nasal gels. This is a triblock copolymer comprised of two blocks one of which is polyethylene oxide (PEO), and the other polypropylene oxide (PPO). The Poloxamer 407 at low temperature is in liquid state due to predominance of the hydrophilic PEO blocks [1]. Nevertheless, at the physiological temperature, the PPO blocks switch to become hydrophobic, which promotes micelle formation and gelation. This property makes it particularly relevant to nasal formulations where gelation should occur as soon as the formulation comes into contact with mucosal tissue.

Poloxamer 407 is relatively inert, non-irritating, non-sensitizing, and suitable for controlled drug delivery of both hydrophilic and lipophilic agents [13].

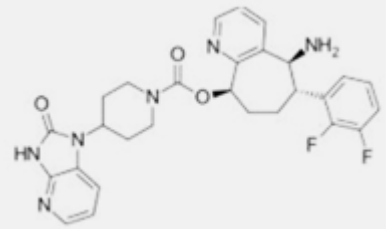
Carbopol is an acrylic polymer accumulated extensively as the pH sensitive ingredient in situ nasal gels. This polymer is soluble in the acidic solutions and turns out to be insoluble when exposed to the pH of the nasal cavity that is, between the range of neutral to slightly acidic. This leads to formation of a highly swollen hydrogel network capable of maintaining drug concentration for lengthy period and release in a controlled manner. Carbopol is prized for its high mucoadhesive, which extends the contact time of the gel to the nasal mucosa, leading to improved drug diffusion. It is frequently combined with other polymers to enhance the combinational effects on gel properties such as stability and drug release property using polymers like Poloxamer.

Chitosan, a natural polymer derived from chitin, is another important component of in-situ nasal gels. It is soluble in acidic conditions and forms a gel when neutralized. Apart from its gelling properties, chitosan is known for its exceptional mucoadhesive ability, which prolongs the retention of the formulation on the nasal mucosa. Furthermore, chitosan has the unique ability to transiently open tight junctions in the nasal epithelium, enhancing drug permeation. This property makes it especially useful for the delivery of large molecules like peptides and proteins. Chitosan is biocompatible, biodegradable, and non-toxic, making it an excellent candidate for nasal drug delivery [14].

Gellan gum is a moisture stabilizing anionic polysaccharide gel forming agent which gels in the presence of divalent cations for example calcium ions. This ion-sensitive polymer can be well applied in nasal gels since gelation of the polymer occurs in response to ionic content of nasal secretion [15]. Gellan gum develops a stable and mechanically strong gel matrix because 5 of ionic crosslinking which fixates the drug and offers controlled release at the same time. Because of its biocompatibility and inherent capability of forming gel in the nasal milieu, the polymer is most suited for long term drug delivery [16]. Sodium alginate is another ion-sensitive polymer commonly used in *in-situ* nasal gel formulations. Extruded from brown seaweed, this substance forms a gel in the calcium ion environment which is inherent in nasal discharge. Calcium ions cause the forming of stable gel when interacted with alginate chains and it interacts well with the nasal mucosa. A second reason for using sodium alginate is that it is highly biocompatible and offers good mucoadhesive properties which prolong the residence time of the formulation and enables a controlled release of the drug. It is particularly advantageous to those products whose active ingredients need to be slowly released and remain active for an extended period [1].

**Drug Profile:**

**TABLE 1: DRUG PROFILE OF RIMEGEPANT**

| | |
|---|---|
| IUPAC Name | [(5S,6S,9R)-5-amino-6-(2,3-difluorophenyl)-6,7,8,9- tetrahydro-5H-cyclohepta[b]pyridin-9-yl] 4-(2-oxo-3H-imidazo[4,5-b]pyridin-1-yl)piperidine-1-carboxylate |
| Description | Rimegepant is a white to off-white powder that belongs to the class of calcitonin gene- related peptide (CGRP) receptor antagonists. It is a small molecule drug with a Molecular weight of 696.56g/mol. |
| Molecular Formula | $C_{28}H_{28}F_2N_6O_3$ |
| Molecular weight | 534.6 |
| Melting point | 175.53°C |
| Chemical Structure |  |

## MATERIALS AND METHODS:

### Experimental:

**Materials:** A sample of Rimegepant API was purchased from Sciquaint Innovations OPC Private Limited, Pune, India. Poloxamer 407, Gellan Gum, Phosphate Buffer pH 7, and Ethanol were acquired from Neeta Chemicals, Pune, India.

**Instruments:** The instruments used for the experimental work were Digital weighing balance, Hot air oven, UV-Visible Spectrophotometer, Fourier Transform Infrared Spectrophotometer, Franz diffusion cell apparatus, Differential

scanning colorimetry (DSC), Water bath (electronic), Digital pH meter, Brookfield Viscometer, Centrifuge machine, and Mucoadhesive strength apparatus.

**Preformulation Study:**

**Organoleptic Characteristics of Drug:** The organoleptic characteristics of the Rimegepant drug substance were determined such as texture, color, odour as well as physical appearance were analyzed and quantified on-site at the laboratory.

**Scanning Absorbance Maxima (λmax):** A 10 µg/ml solution of Rimegepant was scanned across the wavelength range of 200–400 nm. The results showed significant absorbance at 278 nm, where a sharp and prominent peak was observed. Based on this, 278 nm was selected as the detection wavelength in phosphate buffer. The spectral analysis confirmed that Rimegepant exhibits a distinct λmax at 278 nm [17].

**FTIR Spectroscopy:** FTIR spectra of the pure drug were recorded using an FTIR-8400S spectrophotometer (Shimadzu, Japan). To prepare the sample, the drug was finely mixed with potassium bromide (KBr) in a 1:100 ratio using a mortar and pestle. The mixture was then compressed into a pellet using a pellet press, applying a hydraulic pressure of 15 tons for one minute. After compression, the pressure was released by turning the side valve counter clockwise, allowing the pellet to be removed from the die. The formed pellet was placed in the sample holder, and the spectral scan was carried out in the range of 4000 to 400 $cm^{-1}$, with a resolution of 4 $cm^{-1}$ and a scan speed of 2 mm/sec.

**DSC Analysis:** Differential Scanning Calorimetry (DSC) analysis of the pure drug was carried out using a Perkin-Elmer Pyris-1 instrument (Osaka, Japan). To eliminate moisture, the samples were pre-heated before analysis. Approximately 3–7 mg of each sample was precisely weighed and placed into a 40 µL aluminum pan, which was then hermetically sealed. Alpha alumina powder was used as a reference material. The thermograms were recorded over a temperature range of 50°C to 300°C, with a heating rate of 20°C per minute, under a continuous flow of inert nitrogen gas at 20 mL/min. The resulting DSC spectra were analyzed to identify exothermic peak positions and to detect any deviations from the standard thermograms [18].

**Experimental Design:** $3^2$ Factorial design was used to study the effect of two factors on two responses. The factors studied were the amount of Poloxamer 407 and amount of Gellan Gum. The responses studied were Gelation temperature (oC) and Mucoadhesive strength (dyne/$cm^2$). The experimental runs were carried out in randomized order, and the results were analyzed using Design Expert software v.13.0. The layout of the design and actual values used for each factor are presented in **Table 2** [19, 20].

**TABLE 2: LAYOUT OF CENTRAL COMPOSITE DESIGN**

| Independent Variables | | | | | | |
|---|---|---|---|---|---|---|
| **Factors** | **Coded Values** | | | **Actual Values in (%w/v)** | | |
| X1- Poloxamer 407(%w/v) | -1 | 0 | +1 | 18 | 20 | 22 |
| X2-Gellan Gum (%w/v) | -1 | 0 | +1 | 0.2 | 0.3 | 0.4 |
| **Responses (Dependent variable)** | | | | | | |
| Y1 = Gelation temperature ($^oC$) | | | | | | |
| Y2 = Mucoadhesive strength (dyne/$cm^2$) | | | | | | |

## Composition of *In-situ* Nasal Gel:

**TABLE 3: COMPOSITION OF VARIOUS BATCHES OF *IN-SITU* NASAL GEL PREPARED USING 32 FACTORIAL DESIGN**

| F. Code | KF1 | KF2 | KF3 | KF4 | KF5 | KF6 | KF7 | KF8 | KF9 |
|---|---|---|---|---|---|---|---|---|---|
| Rimegepant (mg) | 75 | 75 | 75 | 75 | 75 | 75 | 75 | 75 | 75 |
| Poloxamer 407 (%w/v) | 18 | 20 | 22 | 18 | 20 | 22 | 18 | 20 | 22 |
| Gellan Gum (%w/v) | 0.2 | 0.2 | 0.2 | 0.3 | 0.3 | 0.3 | 0.4 | 0.4 | 0.4 |
| Distilledwater (%w/w) | q.s.to100 | q.s.to100 | q.s.to100 | q.s.to100 | q.s.to100 | q.s.to100 | q.s.to100 | q.s.to100 | q.s.to100 |

**Preparation of *In-situ* Nasal Gel:** The *in-situ* nasal gel was prepared using the cold method. Rimegepant was dissolved in a pre-measured quantity of cold distilled water under constant stirring until completely dissolved. The solution was then refrigerated at 4°C overnight to ensure uniform dissolution. Poloxamer 407, at the specified (18-22 % w/v) concentration, was gradually added to the cold solution with continuous stirring to achieve a homogenous dispersion. Gellan Gum (0.2 – 0.4 % w/v), at the required % w/v concentration, was slowly incorporated into the mixture under constant stirring to ensure uniform distribution. The formulation was refrigerated to allow complete hydration and uniformity. Finally, the volume was adjusted with cold distilled water to achieve the desired composition [21, 22].

**Characterization of *In-situ* Nasal Gel Formulation:**

**i. pH:** The pH of the prepared formulations was measured using universal pH indicator strips ranging from 0 to 14. Each strip was fully dipped into the liquid sample, and the resulting color change was matched with the reference scale provided on the strip packaging [23].

**ii. Gelation Temperature:** The gelation temperature and gelation time of each formulation were evaluated by transferring the formulation into a test tube and placing it in a water bath maintained at 15°C. The temperature was then gradually increased at a rate of 1°C per minute. The test tube was tilted to a 180° angle every minute, and the temperature at which the formulation stopped flowing, indicating a phase transition, was recorded as the gelation temperature. The time taken for gelation was also noted. The formulation with a gelation temperature closest to the nasal cavity temperature (32–34°C) was selected as the optimal formulation [17].

**iii. Viscosity:** The viscosity of the *in-situ* gel formulations was measured using a Brookfield DVII+ Pro viscometer equipped with an S-94 spindle (Ametek Brookfield Viscometers, USA). The prepared gels were placed into a beaker, and the spindle was immersed vertically into the sample. Measurements were taken at 100 rpm while maintaining the temperature at 37±0.5°C. Viscosity readings were recorded as the system cooled, and all measurements were carried out in triplicate [17].

**iv. Drug Content Assay:** 1 ml of the prepared formulation was added to 10 ml of phosphate buffer and mixed for 2–3 minutes with occasional shaking. The resulting solution was then filtered using a 0.45 µm filter paper and further diluted with phosphate buffer. The concentration of Rimegepant in the formulation was quantified spectrophotometrically at 278 nm using a Shimadzu 1800 UV-Visible spectrophotometer (Japan) [24].

**v. Spreadability:** The spreadability of the formulation was evaluated using the parallel-plate method. One gram of the sample, prepared 48 hours prior to testing, was placed between two glass plates measuring 20 x 20 cm. A weight of 125 g was applied on the top plate for 1 minute. Afterward, the diameter of the spread sample between the plates was measured25.

**vi. *Ex-vivo* Drug Permeation Studies and Mucoadhesive Strength Including Flux and KP:** *Ex-vivo* drug permeation of the Rimegepant-loaded thermosensitive in-situ nasal gel was assessed using a Franz diffusion cell. Freshly prepared nasal mucosa was positioned between the donor and receptor compartments of the cell. The receptor compartment was filled with phosphate buffer (pH 7) maintained at 37°C and stirred continuously at 100 rpm. A 10 mg portion of the gel was applied onto the mucosa, and the compartments were securely clamped together. At specific time intervals (0, 0.5, 1, 2, 4, 6, 8, 10, and 12 hours), 1 ml samples were withdrawn from the receptor compartment and immediately replaced with fresh phosphate buffer (pH 7) to maintain sink conditions. The withdrawn samples were filtered through a 0.45 µm membrane, appropriately diluted, and analyzed for drug content at 278 nm using a Shimadzu 1800 UV-Visible spectrophotometer (Japan). To understand the drug release mechanism, the permeation data were fitted into various kinetic models zero-order, first- order, Higuchi, and Korsmeyer-Peppas. The release rate constants (k) and diffusion exponent (n) were determined through linear regression using Microsoft Excel 365. The coefficient of

determination ($R^2$) was used to assess the goodness of fit for each model [26].

**vii. Stability Studies:** The stability of the selected in-situ gel formulation was evaluated through a series of stress tests, including six heating-cooling cycles between 4°C and 40°C, three freeze-thaw cycles between −21°C and 25°C, and centrifugation at 13,000 rpm for 30 minutes. Following each test, the samples were visually inspected for signs of instability such as changes in clarity, physical appearance, texture, and other noticeable alterations [27].

**Pre-formulation Study:**

**Organoleptic Properties:** The physicochemical characterization of Rimegepant yielded comprehensive insights into its properties, which are crucial for developing an effective thermoreversible in-situ nasal gel formulation. The organoleptic evaluation presented in **Table 4** confirmed that Rimegepant appears as a white to off-white crystalline powder with a pleasant odour, aligning perfectly with the reported standard specifications. These characteristics are favorable for nasal drug delivery as they suggest good patient acceptability.

## RESULTS AND DISCUSSION:

**TABLE 4: RESULTS OF ORGANOLEPTIC PROPERTIES**

| Sr. no. | Parameter | Observed Value | Reported Value |
|---|---|---|---|
| 1 | Color | White to off-white | White to off-white |
| 2 | Odor | Pleasant | Pleasant |
| 3 | Texture | Crystalline powder | Crystalline powder |

**Scanning Absorbance Maxima (λmax):** The UV spectrophotometric analysis of Rimegepant, as shown in **Fig. 1**, revealed the maximum wavelength of absorption (λmax) at 278.4 nm, which was subsequently used for quantitative analysis. The amount of Rimegepant in the formulation was determined spectrophotometrically at 278nm as depicted in **Fig. 2.**

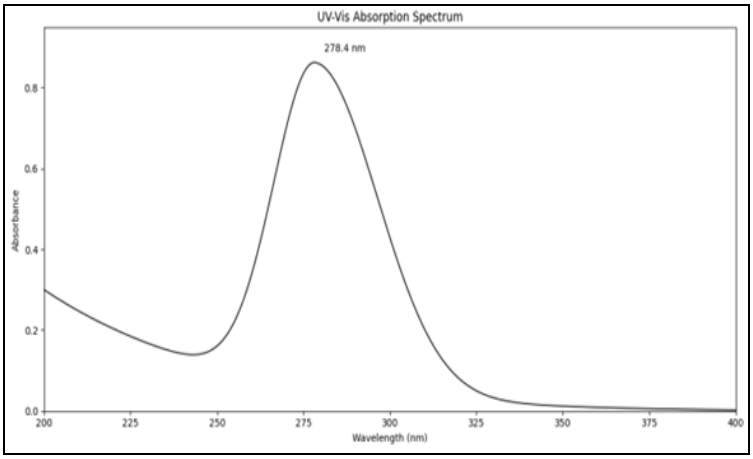



**FIG. 1: UV SCANNING ABSORBANCE MAXIMA (ΛMAX) OF RIMEGEPANT**

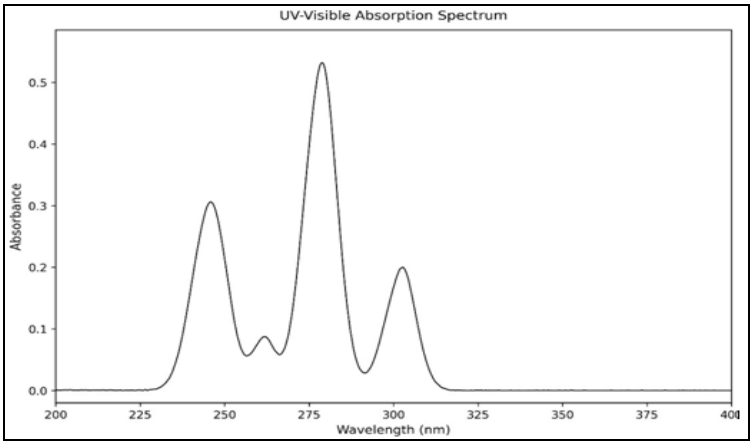



**FIG. 2: UV SCANNING ABSORBANCE MAXIMA (ΛMAX) OF RIMEGEPANT IN FORMULATION**

**FTIR Spectroscopy:** The FTIR spectroscopic analysis provided detailed information about the structural integrity of Rimegepant and potential molecular interactions in the physical mixture. The FTIR spectrum of pure Rimegepant and its physical mixture (Poloxamer 407 and Gellan gum) **Fig. 3** and **4** exhibited characteristic peaks that align with its molecular structure.

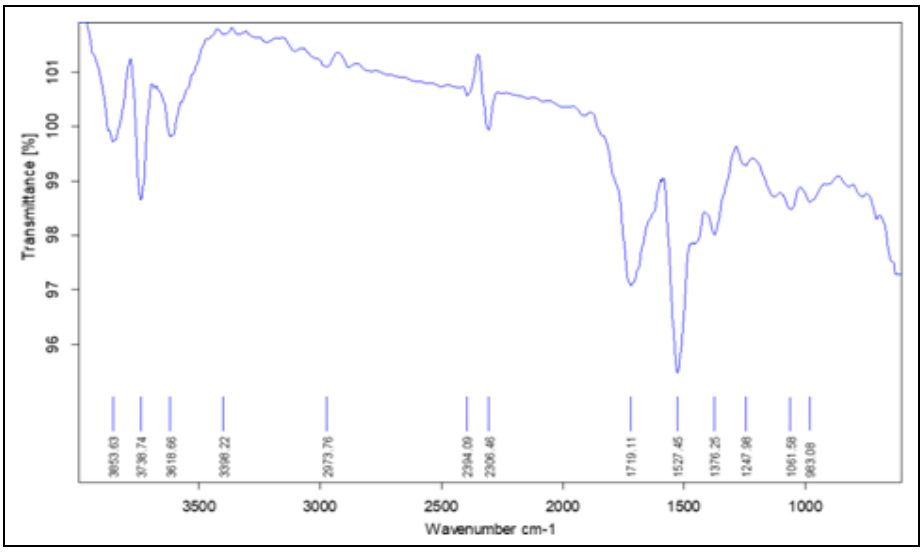



**FIG. 3: FTIR SPECTRA OF RIMEGEPANT**

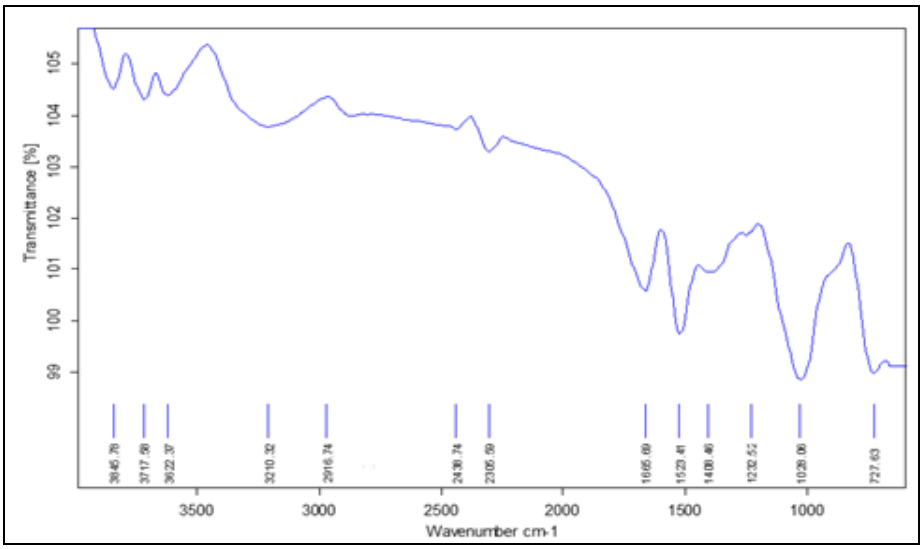



**FIG. 4: FTIR SPECTRA OF PHYSICAL MIXTURE (RIMEGEPANT + POLOXAMER 407 + GELLAN GUM)**

**TABLE 5: INTERPRETATION OF FTIR RESULTS**

| Functional Group | Rimegepant ($cm^{-1}$) | Physical Mixture ($cm^{-1}$) |
|---|---|---|
| N-H stretching (primary amine) | 3853.63,3738.74 | 3845.78,3717.58 |
| N-H stretching (secondary) | 3398.22 | 3210.32 |
| C-H stretching | 2973.76 | 2916.74 |
| C ≡ N stretching | 2394.09,2306.46 | 2438.74,2305.59 |
| C=O stretching | 1719.11 | 1665.69 |
| C=C aromatic | 1527.45 | 1523.41 |
| C-H bending | 1376.35 | 1408.46 |
| C-N stretching (aromatic amine) | 1247.98 | 1232.52 |
| C-F stretching | 1061.58 | 1028.28 |
| C-H out-of-plane | 983.08 | 727.63 |
| O-H stretching (P407) | - | 3622.37 |

**DSC Analysis:** The DSC analysis, illustrated in **Fig. 5**, showed a sharp endothermic peak for Rimegepant at 175.53°C, corresponding to its melting point. This sharp peak indicates the

crystalline nature of the drug and its high purity. When examining the physical mixture's DSC spectra **Fig. 6**, a similar endothermic peak was observed at 175.71°C, with only a minimal shift of 0.18°C from the pure drug's melting point. This negligible shift suggests excellent compatibility between Rimegepant and the selected excipients, as significant interactions would have resulted in more substantial peak shifts or the appearance of new thermal events.

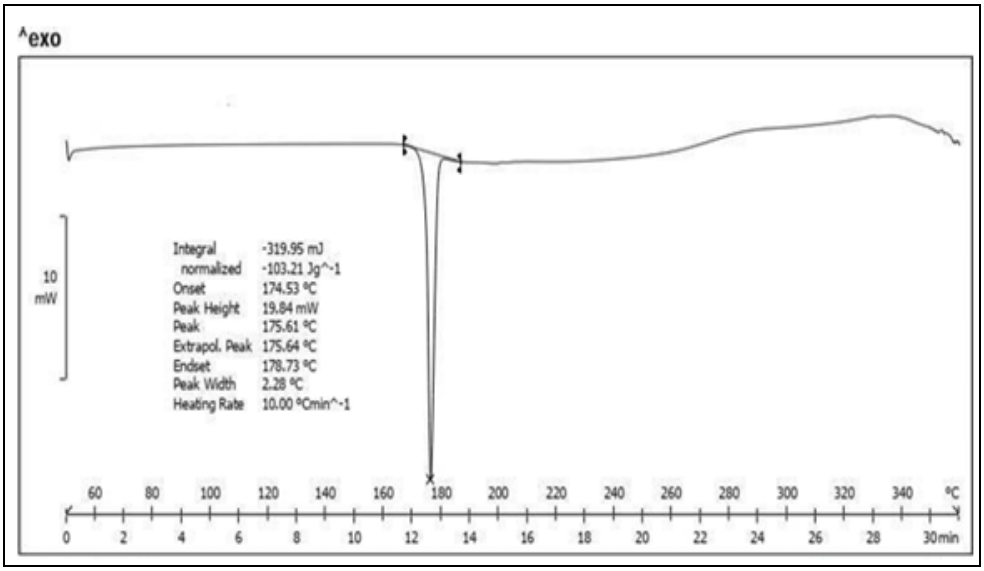



**FIG. 5: DSC SPECTRA OF PURE RIMEGEPANT (175.53ºC)**

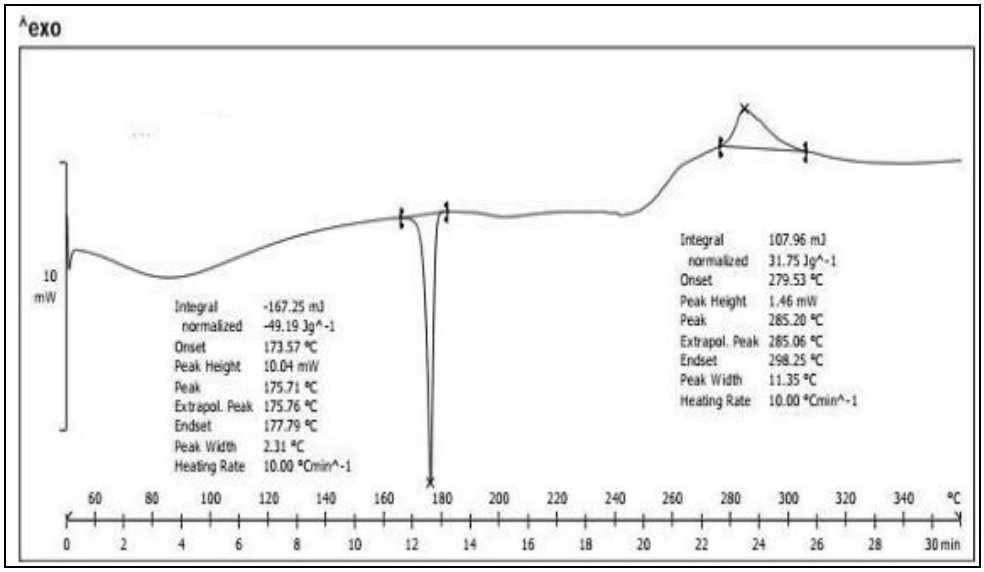



**FIG. 6: DSC SPECTRA OF PHYSICAL MIXTURE OF RIMEGEPANT + POLOXAMER 407 + GELLAN GUM (175.71ºC)**

**Evaluation of *In-situ* Nasal Gel:** The thermosensitive properties of the formulations, presented in **Table 6**, showed varying gelation temperatures ranging from 22.52±0.38°C to 42.12±0.58°C. Notably, formulation KF9 exhibited the lowest gelation temperature (22.52±0.38°C) and the shortest gelation time (6.45±2.3 seconds), coupled with the highest viscosity (142.12±3.73 m.Pa. s). In contrast, KF4 showed the highest gelation temperature (42.12±0.58°C).

**TABLE 6: CHARACTERIZATION OF THERMOSENSITIVE *IN-SITU* NASAL GEL FORMULATIONS**

| Formulation | Gelation temperature (°C) | Gelation time (sec) | Viscosity (m.Pa.s) |
|---|---|---|---|
| KF1 | 40.68±0.45 | 21.45±2.4 | 45.12±1.85 |
| KF2 | 36.31±0.34 | 8.68±1.1 | 46.76±0.78 |
| KF3 | 24.45±0.28 | 13.34±2.2 | 123.34±1.62 |

| | | | |
|---|---|---|---|
| KF4 | 42.12±0.58 | 8.23±1.5 | 47.89±2.93 |
| KF5 | 33.89±0.43 | 34.56±1.8 | 49.67±0.84 |
| KF6 | 26.76±0.31 | 11.89±1.2 | 98.43±0.71 |
| KF7 | 39.45±0.57 | 18.12±3.6 | 48.98±1.89 |
| KF8 | 35.23±0.45 | 9.76±1.4 | 51.76±0.96 |

Results are shown in mean ± SD, (n=3).

**Mucoadhesive Strength and Spreadability:** The mucoadhesive strength and spreadability results, documented in **Table 7,** provide insights into the formulations' ability to adhere to and spread across the nasal mucosa. Formulation KF8 demonstrated the highest mucoadhesive strength (6972.2±19.39 dyne/cm²), followed by KF5 (6621.7±18.48 dyne/cm²), suggesting superior retention properties. The spreadability values ranged from 6.9±0.74 cm to 12.3±1.49 cm, with KF2 showing the highest spreadability (12.3±1.49 cm). These parameters are particularly important as they influence the residence time of the formulation in the nasal cavity and its ability to cover the absorption surface effectively.

**TABLE 7: MUCOADHESIVE STRENGTH AND SPREADABILITY OF THERMOSENSITIVE *IN-SITU* NASAL GEL FORMULATIONS**

| Formulation | Mucoadhesive strength (dyne/cm$^2$) | Spreadability (cm) |
|---|---|---|
| KF1 | 5232.1±5.31 | 7.8±0.67 |
| KF2 | 5673.4±8.78 | 12.3±1.49 |
| KF3 | 4329.9±13.83 | 9.8±0.59 |
| KF4 | 5724.5±9.53 | 8.9±0.73 |
| KF5 | 6621.7±18.48 | 10.7±1.43 |
| KF6 | 5129.3±8.82 | 9.7±0.39 |
| KF7 | 5723.8±12.68 | 6.9±0.74 |
| KF8 | 6972.2±19.39 | 8.7±0.52 |
| KF9 | 5789.2±13.53 | 10.8±0.93 |

Results are shown in mean ± SD, (n=3).

**Optimization of Formulation:**

**For Gelation temperature (R1):**

**ANOVA for Quadratic model for Gelation Temperature (R1):** The ANOVA results show that the quadratic model for gelation temperature is statistically significant (p = 0.0019). Poloxamer 407 and its quadratic term (A²) significantly affect gelation temperature, while Gellan Gum, the interaction term (AB), and B² are not significant. This indicates that Poloxamer 407 plays a major role in determining the gelation behavior of the formulation.

**TABLE 8: ANOVA FOR THE QUADRATIC MODEL FOR GELATION TEMPERATURE (R1)**

| Source | Sum of Squares | df | Mean Square | F-value | p-value | |
|---|---|---|---|---|---|---|
| Model | 423.71 | 5 | 84.74 | 88.28 | 0.0019 | significant |
| A-Poloxamer407 | 392.37 | 1 | 392.37 | 408.75 | 0.0003 | |
| B-GellanGum | 3.00 | 1 | 3.00 | 3.12 | 0.1754 | |
| AB | 0.1225 | 1 | 0.1225 | 0.1276 | 0.7446 | |
| A² | 20.95 | 1 | 20.95 | 21.83 | 0.0185 | |
| B² | 7.27 | 1 | 7.27 | 7.57 | 0.0706 | |
| Residual | 2.88 | 3 | 0.9599 | | | |
| CorTotal | 426.59 | 8 | | | | |

The regression equation obtained for Gelation temperature is as follows: Gelation temperature = +7.31 -0.428*A -0.466*B +1.08*AB +2.53A2+0.965*B2

**Method Operable Design Region (MODR):** The contour plot shows that increasing Poloxamer 407 concentration significantly decreases gelation temperature, as indicated by the colour shift from red to blue. Gellan Gum has a comparatively minor effect, with minimal changes along the Y-axis. This confirms that Poloxamer 407 is the key factor influencing gelation temperature.

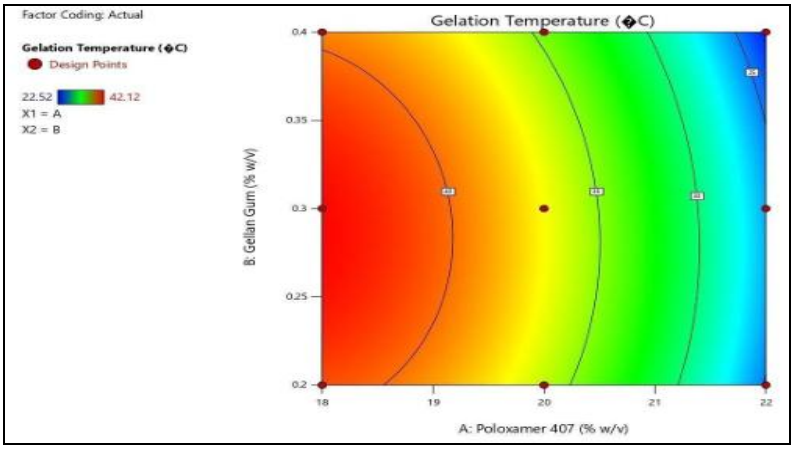



**FIG. 7: CONTOUR PLOT SHOWING EFFECT OF POLOXAMER 407 AND GELLAN GUM ON GELATION TEMPERATURE (R1)**

**3D Response Surface Plot:** The 3D response surface plot visually demonstrates the effect of Poloxamer 407 and Gellan Gum concentrations on gelation temperature (R1). The surface shows a steep decline in gelation temperature with increasing Poloxamer 407 concentration, confirming its strong influence. In contrast, changes in Gellan Gum concentration have a milder effect, as indicated by the gentle slope along the B-axis.

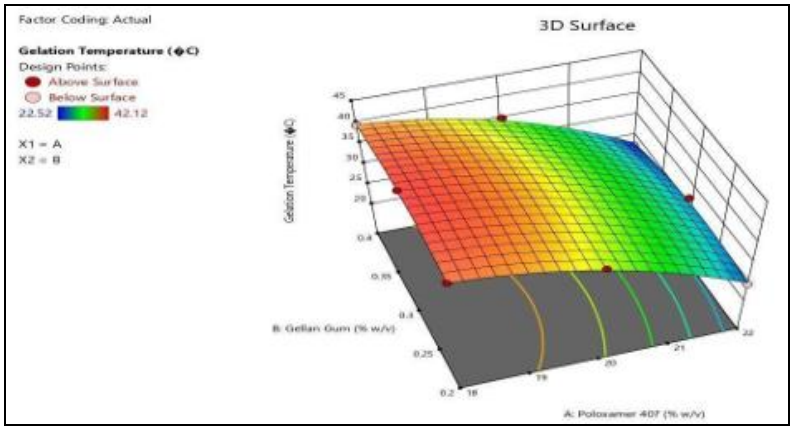



**FIG. 8: 3D RESPONSE SURFACE PLOT SHOWING EFFECT OF POLOXAMER 407 AND GELLAN GUM ON GELATION TEMPERATURE (R1)**

**Perturbation Plot:** The perturbation plot shows that gelation temperature is highly sensitive to changes in Poloxamer 407 concentration, as indicated by the steep green curve. In contrast, Gellan Gum has a milder effect, shown by the flatter blue line. This confirms Poloxamer 407 as the dominant factor affecting gelation temperature.

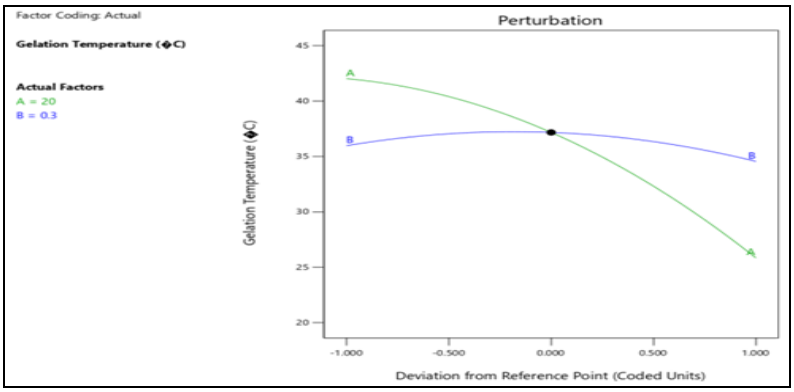



**FIG. 9: PERTURBATION PLOT SHOWING EFFECT OF POLOXAMER 407 AND GELLAN GUM ON GELATION TEMPERATURE (R1)**

**For Mucoadhesive Strength (R2):**

**ANOVA for Quadratic model for Mucoadhesive Strength (R2):** The ANOVA table for mucoadhesive strength (R2) shows the model is statistically significant (p = 0.0039, F = 53.76). Both Poloxamer 407 and Gellan Gum significantly influence the response, with Gellan Gum showing a stronger effect. The interaction (AB) and quadratic terms, particularly $A^2$, are also significant contributors. The low residual value indicates a good model fit.

**TABLE 9: ANOVA FOR THE QUADRATIC MODEL MUCOADHESIVE STRENGTH (R2)**

| Source | Sum of Squares | df | Mean Square | F-value | p-value | |
|---|---|---|---|---|---|---|
| Model | 4.844E+06 | 5 | 9.689E+05 | 53.76 | 0.0039 | significant |

The regression equation obtained for Mucoadhesive strength is as follows: Mucoadhesive strength = +7.31 -0.428*A -0.466*B +1.08*AB +2.53A2+0.965*B2

**Method Operable Design Region (MODR):** The contour plot demonstrates the interactive effect of Poloxamer 407 and Gellan Gum concentrations on mucoadhesive strength (R2). Maximum mucoadhesive strength is observed around the mid-range concentration of Poloxamer 407 (approximately 20% w/v) and higher levels of Gellan Gum (around 0.35–0.4% w/v), as indicated by the red-orange region. At both lower and higher concentrations of Poloxamer 407, the mucoadhesive strength decreases, reflecting a parabolic relationship.

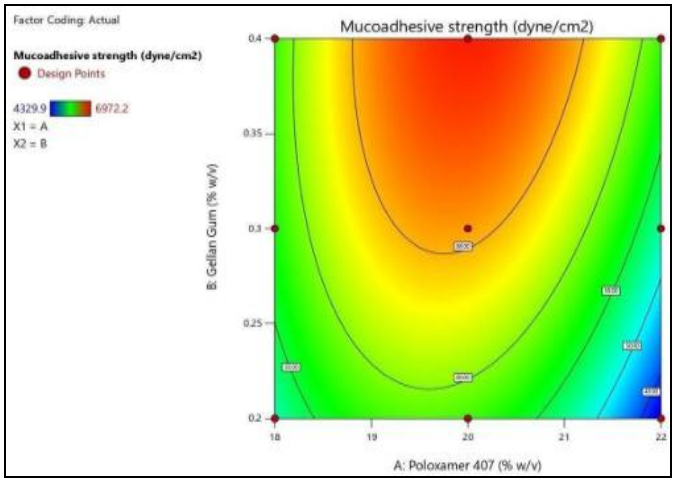



**FIG. 10: CONTOUR PLOT SHOWING EFFECT OF POLOXAMER 407 AND GELLAN GUM ON MUCOADHESIVE STRENGTH (R2)**

**3D Response Surface Plot:** The 3D response surface plot reveals the interactive effect of Poloxamer 407 and Gellan Gum concentrations on mucoadhesive strength (R2).

The plot shows that mucoadhesive strength increases with increasing Gellan Gum concentration and reaches a peak at an intermediate level of Poloxamer 407 (around 20% w/v), beyond which the strength begins to decline. This curved surface indicates significant quadratic effects, especially for Poloxamer 407, consistent with the ANOVA results. The highest mucoadhesive strength is observed when both components are optimally balanced, highlighting their synergistic interaction.

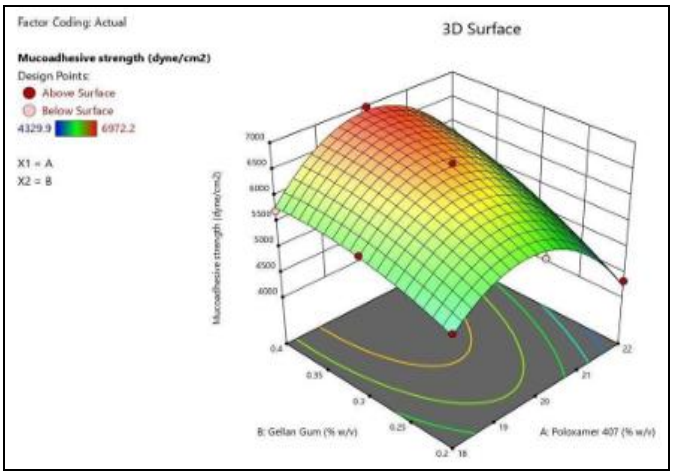



**FIG. 11: 3D RESPONSE SURFACE PLOT SHOWING EFFECT OF POLOXAMER 407 AND GELLAN GUM ON MUCOADHESIVE STRENGTH (R2)**

**Pertubation Plot:** The perturbation plot illustrates the sensitivity of mucoadhesive strength (R2) to variations in Poloxamer 407 (A) and Gellan Gum (B) from a central reference point.

The curved green line representing Poloxamer 407 shows a pronounced peak, indicating that small deviations around the optimal concentration significantly affect mucoadhesive strength, with both higher and lower concentrations reducing it.

In contrast, the blue line for Gellan Gum is more linear and gradually rising, suggesting a consistent and milder positive effect. Overall, Poloxamer 407 has a stronger and more non-linear impact on mucoadhesive strength compared to Gellan Gum.

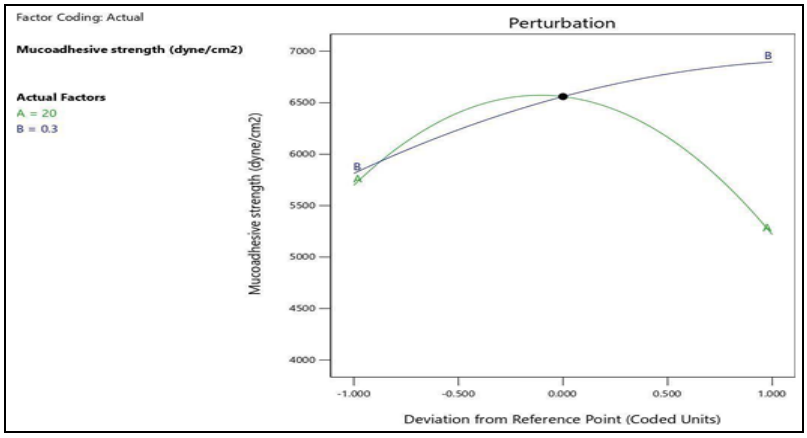



**FIG. 12: PERTURBATION PLOT SHOWING EFFECT OF POLOXAMER 407 AND GELLAN GUM ON MUCOADHESIVE STRENGTH (R2)**

***Ex-vivo* Permeation Studies of Thermosensitive *In-situ* Nasal Gel Using Goat Nasal Mucosa:** The *ex-vivo* permeation studies conducted on goat nasal mucosa provided crucial insights into the drug release behaviour of the thermoreversible *in-situ* nasal gel formulations over a 10-hour period, as presented in **Table 10** and visualized in **Fig. 13**. The permeation profiles demonstrated significant variations among the nine formulations (KF1-KF9), with KF8 showing the most promising drug release pattern.

KF8 exhibited a well-controlled initial release of 13.02±0.65% at 1 hour, followed by sustained release reaching 98.31±4.92% at 10 hours, indicating optimal drug permeation characteristics.

**TABLE 10: *EX-VIVO* DRUG PERMEATION PROFILE OF RIMEGEPANT FROM DIFFERENT THERMOSENSITIVE *IN-SITU* NASAL GEL FORMULATIONS (KF1-KF9) (MEAN ± SD, N=3)**

| 10 | 9 | 8 | 7 | 6 | 5 | 4 | 3 | 2 | 1 | Time (hours) |
|---|---|---|---|---|---|---|---|---|---|---|
| - | - | 95.48± 4.77 | 84.06±4 .20 | 74.09± 3.70 | 63.41± 3.17 | 51.22±2 .56 | 38.79± 1.94 | 25.48± 1.27 | 12.53±0 .63 | KF1 |
| 74.50±3.73 | 69.20±3 .46 | 61.43± 3.07 | 43.79±2 .19 | 37.57± 1.88 | 31.28±1 .56 | 25.32±1 .27 | 18.70± 0.94 | 12.50± 0.63 | 6.21±0. 31 | KF2 |
| 80.42±4.02 | 74.89±3 .74 | 67.54± 3.38 | 60.59±3 .03 | 53.43± 2.67 | 45.36±2 .27 | 36.58±1 .83 | 27.83± 1.39 | 18.52± 0.93 | 9.26±0. 46 | KF3 |
| 78.54±3.93 | 72.52±3 .63 | 66.91± 3.35 | 59.14±2 .96 | 51.52± 2.58 | 43.59±2 .18 | 35.56±1 .78 | 26.43± 1.32 | 17.62± 0.88 | 8.71±0. 44 | KF4 |
| 96.61±4.83 | 91.32±4 .57 | 83.14± 4.16 | 74.37±3 .72 | 66.87± 3.34 | 56.53±2 .83 | 46.17±2 .31 | 35.09± 1.75 | 23.51± 1.18 | 11.04± 0.55 | KF5 |
| 84.11±4.21 | 70.54±3 .53 | 64.29± 3.21 | 51.22±2 .56 | 44.58± 2.23 | 37.51±1 .88 | 30.23±1 .51 | 22.82± 1.14 | 15.39± 0.77 | 7.52±0. 38 | KF6 |
| 94.32±4.72 | 88.45±4 .42 | 81.21± 4.06 | 73.24±3 .66 | 64.29± 3.21 | 54.51±2 .73 | 44.36±2 .22 | 33.52± 1.68 | 22.14± 1.11 | 10.82±0 .54 | KF7 |
| 98.31±4.92 | 95.78±4 .79 | 89.14± 4.46 | 81.07±4 .05 | 72.08± 3.60 | 62.19±3 .11 | 51.17±2 .56 | 39.09± 1.95 | 26.52± 1.33 | 13.02±0 .65 | KF8 |
| - | 83.31±4 .17 | 75.53± 3.78 | 66.78±3 .34 | 58.30± 2.92 | 50.04±2 .50 | 41.73±2 .09 | 33.31± 1.67 | 25.89± 1.29 | 16.72±0 .84 | KF9 |

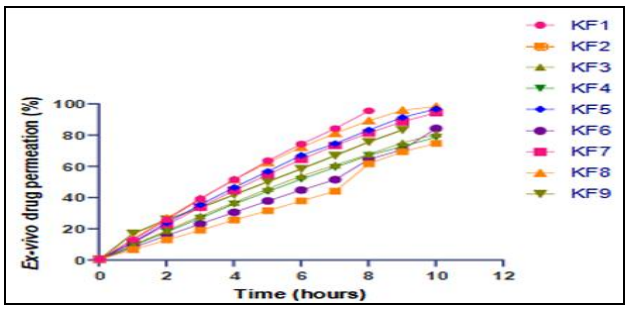



**FIG. 13: *EX-VIVO* DRUG PERMEATION STUDIES OF FORMULATIONS ON GOAT NASAL MUCOSA (KF1-KF9)**

**Flux and Kp:** Flux and the permeability coefficient (Kp) in formulation, are determined through in vitro diffusion studies, using a modified Franz diffusion cell. Flux (J) measures the amount of drug passing through a membrane per unit area and time. Kp, the permeability coefficient, is calculated using the flux and the concentration gradient across the membrane.

**TABLE 11: FLUX AND KP OF THERMOSENSITIVE *IN-SITU* NASAL GEL FORMULATIONS (KF1- KF9)**

| Batch | Flux (µg/cm²/h) | Kp (cm/h) |
|---|---|---|
| KF1 | 12.76 | 0.00170 |
| KF2 | 6.28 | 0.00084 |
| KF3 | 9.09 | 0.00121 |
| KF4 | 8.78 | 0.00117 |
| KF5 | 11.42 | 0.00152 |
| KF6 | 7.52 | 0.00100 |
| KF7 | 10.99 | 0.00146 |

**Stability Studies:** The stability study of the optimized formulation KF8, conducted over six months at 25°C ± 2°C/60% ± 5% RH **Table 12**, demonstrated remarkable physicochemical stability. The formulation maintained its clarity and transparent appearance throughout the study period. Critical parameters such as pH remained constant at 6.1, indicating minimal chemical degradation. The physical parameters showed minimal variations: spreadability ranged from 10.8±0.13 to 11.8±0.78 cm, viscosity remained stable between 51.48±1.89 and 52.93±0.28 cps, and mucoadhesive strength showed only a slight decrease from 6974±0.673 to 6838±0.982 dyne/cm². The thermoreversible properties remained consistent, with gelation time and temperature showing minimal variations (9.76±0.41 to 9.61±0.74 seconds and 35.83±0.78 to 35.44±0.36°C, respectively). Most importantly, the ex-vivo permeation capability at 10 hours remained high, decreasing only slightly from 98.78±0.47% to 97.11±0.96% over the six-month period.

**TABLE 12: PHYSICOCHEMICAL STABILITY ASSESSMENT OF OPTIMIZED RIMEGEPANT *IN-SITU* NASAL GEL FORMULATION (KF8) STORED AT 25°C ± 2°C/60% ± 5% RH OVER A SIX-MONTH PERIOD**

| Response | 0 month | 1 month | 3 months | 6 months |
|---|---|---|---|---|
| Clarity | Clear | Clear | Clear | Clear |
| Visual appearance | Transparent | Transparent | Transparent | Transparent |
| pH | 6.1±0.06 | 6.1±0.06 | 6.1±0.09 | 6.1±0.02 |
| Spread ability (cm) | 11.1±0.92 | 11.6±0.42 | 11.8±0.78 | 10.8±0.13 |
| Viscosity (cps) | 51.48±1.89 | 51.69±0.42 | 51.84±1.57 | 52.93±0.28 |
| Mucoadhesive strength (dyne/cm$^2$) | 6974±0.673 | 6953±0.675 | 6851±0.872 | 6838±0.982 |
| Gelationtime (sec) | 9.76±0.41 | 9.67±0.32 | 9.66±0.81 | 9.61±0.74 |
| Gelation Temperature | 35.83±0.78 | 35.52±0.67 | 35.51±0.72 | 35.44±0.36 |

**Future Perspectives:** *In-situ* nasal gels, which form a gel in the nasal cavity after administration, offer a promising future in drug delivery, particularly for systemic and brain- targeted therapies. They allow for easy administration in liquid form, ensuring precise dosing, and prolonged retention due to gel formation. This approach can improve patient compliance and bioavailability, especially for drugs with poor oral bioavailability or those needing to bypass first- pass metabolism.

**CONCLUSION:** The successful development and optimization of Rimegepant loaded thermo-reversible in-situ nasal gel formulation demonstrates a promising approach for enhanced migraine treatment. The systematic evaluation of physicochemical properties, thermal characteristics, and drug-excipient compatibility through DSC and FTIR analyses confirmed the formulation's stability and integrity. The optimized formulation KF8, containing Poloxamer 407 and Gellan Gum, exhibited ideal characteristics including appropriate gelation temperature (35.23°C), rapid gelation time (9.76 seconds), and strong mucoadhesive strength (6972.2 dyne/cm²). The *ex-vivo* permeation studies revealed efficient drug delivery with 98.31% permeation over 10 hours, supported by favourable flux (12.51 µg/cm²/h) and permeability coefficient (0.00167 cm/h) values.

These comprehensive findings suggest that the developed thermoreversible *in-situ* nasal gel formulation offers a viable, patient-friendly alternative for migraine treatment, combining the advantages of ease of administration, improved retention time, and sustained drug release. The successful development and characterization of this novel drug delivery system provide a strong foundation for further clinical investigations and potential commercialization, potentially leading to

improved therapeutic outcomes for migraine patients.

**ACKNOWLEDGEMENT:** Authors are thankful to Principal, Abhinav Education Society's, College of pharmacy, Narhe, Pune.

**CONFLICTS OF INTEREST:** Nil

## REFERENCES:


1. Çelikys, Örenli B, Al-Mohaya M, MESUTB and Özsoy Y: Nasal *in-situ* gels as a drug delivery system: An overview of literature and clinical studies. Journal of Research in Pharmacy 2023; 27.
2. Garg A, Agrawal R, Chauhan CS and Deshmukh R: *In-situ* gel: as mart carrier for drug delivery. International Journal of Pharmaceutics 2024; 123819.
3. Bakhrushina EO, Mikhel JB, Kondratieva VM, Demina NB and Grebennikova TV: Insitugelsas a modern method of intranasal vaccine delivery. Problems of Virology 2022; 67: 395–402.
4. Bai B, Leng J and Wei M: A comprehensive review of in-situ polymer gel simulation for conformance control. Petroleum Science 2022; 19: 189–202.
5. RV K, P G, Mamledesai S, VU K and Aladakatti R: Synergistic drug compatibility of sumatriptansuccinate and metoclopramide hydrochloride (*In-situ* gel formulations) for nasal drug release optimization. Asian Journal of Pharmaceutical and Clinical Research 2024; 17: 132–8. https://doi.org/10.22159/ajpcr.2024v17i3.48485.
6. Alabdly A and Kassab HJ: Formulation variables effect on gelation temperature of nefopam hydrochloride intranasal in situ gel (Conference Paper). Iraqi Journal of Pharmaceutical Sciences (P-ISSN 1683-3597 E-ISSN 2521-3512) 2022; 31: 32–44.
7. Ahirwar G, Chouksey MK and Ganju DK: Formulation and development of nasal *in-situ* gel of rizatriptan for treatment of migraine. Journal of Advanced Zoology 2024; 45: 1307–14. https://doi.org/10.53555/jaz.v45i2.4125.
8. Jadhav D, Shaikh M, Jain H, Sahu A and Meshram DB: Formulation and Evaluation Xylometazoline Hydrochloride Mucoadhesive Nasal *in-situ* Gel 2022.
9. Bakhrushina EO, Mikhel IB, Kondratieva VM, Zubareva IM, Kosenkova SI and Belyatskaya AV: Intranasal Ion-Triggered *in-situ* Delivery System of Virus- like Particles: Development Using the Quality by Design Approach. Polymers 2024; 16: 685.
10. Dewan M, Adhikari A, Jana R and Chattopadhyay D: Development, evaluation and recent progress of ocular in situ gelling drug delivery vehicle based on poloxamer 407. J of Drug Delivery Science and Technology 2023; 104885.
11. Hamdi NAM, Azmi NA, Sabari NHM, Harun AF, Haris MS: An insight into the use and advantages ofCarbopol in topical mucoadhesive drug delivery system: Asystematic review. Journal of Pharmacy 2023; 3:53–65.
12. Amarsing TS and Pandit PS: Development of Nasal *In-situ* Gel Formulation of Fexofenadine HCl Using Gellan Gum (Gelerite®). Interpretation 2023; 1: 1200– 1020.
13. Lavania K and Garg A: Ion-activated *In-Situ* Gel of Gellan Gum Containing Chrysin for Nasal Administration in Parkinson's disease. Recent Advances in Drug Delivery and Formulation: Formerly Recent Patents on Drug Delivery & Formulation 2024; 18: 35–49.
14. Nair AB, Chaudhary S, Shah H, Jacob S, Mewada V and Shinu P: Intranasal Delivery of Darunavir-Loaded Mucoadhesive *in-situ* Gel: Experimental Design, *in-vitro* Evaluation, and Pharmacokinetic Studies. Gels 2022; 8: 342. https://doi.org/10.3390/gels8060342.
15. Jadhav P, Jadhav R, Gondkar S and Bachhav R: An over view on in-Situ nasal gel for Drug Delivery 2023.
16. Kolawole OM and Cook MT: *In-situ* gelling drug delivery systems for topical drug delivery. European Journal of Pharmaceutics and Biopharmaceutics 2023; 184: 36– 49. https://doi.org/10.1016/j.ejpb.2023.01.007.
17. Mahajan HS, Jadhao VD and Chandankar SM: Pullulan and Pluronic F-127 based *in-situ* gel system for intranasal delivery: Development, *in-vitro* and *in-vivo* evaluation. Journal of Bioactive and Compatible Polymers 2022; 37: 406–18. https://doi.org/10.1177/08839115221110284
18. Unger-Manhart N, Morokutti-Kurz M, Zieglmayer P, Russo A, Siegl C and König-Schuster M: Decongestant Effect of" Coldamaris Akut", a Carrageenan- and Sorbitol-Containing Nasal Spray in Seasonal Allergic Rhinitis. IJGM 2024; 17: 5105–21. https://doi.org/10.2147/IJGM.S476707.
19. Bakhrushina E, Mikhel I, Kondratieva V, Demina N, Grebennikova T, Krasnyuk Jr I, et al. Main Aspects of Pharmaceutical Development of In Situ Immunobiological Drugs for Intranasal Administration. Current Pharmaceutical Biotechnology 2024; 25:1394–405.
20. Mankar SD, Parjane SR, Siddheshwar SS and Dighe SB: Formulation, Optimization and *in-vivo* Characterization of Thermosensitive *in-situ* Nasal Gel Loaded with Bacosidea for Treatment of Epilepsy. AAPS Pharm Sci Tech 2024; 25: 151. https://doi.org/10.1208/s12249-024-02870-2.
21. Mohananaidu K, Chatterjee B, Mohamed F, Mahmood S and Hamed Almurisi S: Thermoreversible carbamazepine *in-situ* gel for intranasal delivery: development and *in-vitro*, *ex-vivo* evaluation. AAPS Pharm Sci Tech 2022; 23: 288. https://doi.org/10.1208/s12249-022-02439-x
22. Özkan B, Altuntaş E, Ünlü Ü, Doğan HH, Özsoy Y and Çakır Koç R: Development of an antiviral Ion-activated *In-situ* gelcontaining 18β-glycyrrhetinic acid: A promising alternative against respiratory syncytial virus. Pharmaceutics 2023; 15: 2055.
23. Mali A and Bhanwase A: *In-vitro*, *ex-vivo* & *in-vivo* assessment of brain targeted thermoreversible mucoadhesive *in-situ* intranasal gel of carmustine for the treatment of glioblastoma. Bio Nano Sci 2024; 14: 2571–81. https://doi.org/10.1007/s12668-024-01521-x
24. Bekhet MA, Ali AA, Kharshoum RM, El-Ela FIA and Salem HF: Intranasal niosomal *in-situ* gel as a novel strategy for improving citicoline efficacy and brain delivery in treatment of epilepsy: *in-vitro* and *ex-vivo* characterization and *in-vivo* pharmacodynamics investigation. J of Pharma Sciences 2022; 111: 2258–69.
25. Gadhave D, Khot S, Tupe S, Shinde M, Tagalpallewar A and Gorain B: Nose- to-brain delivery of octreotide acetate *in-situ* gel for pituitary adenoma: pharmacological and *in-vitro* cytotoxicity studies. International Journal of Pharmaceutics 2022; 629: 122372.
26. Prajapati A, Patel J, Kale SU, Kansara BR, Khan NT and Kolte PG: Development of an *in-situ* nasal gel formulation of bepotastine besilate for the treatment of allergic rhinitis. Research Journal of Pharmacy and Technology 2024; 17: 3745– 51.
27. Cunha S, Swedrowska M, Bellahnid Y, Xu Z, Lobo JS and Forbes B: Thermosensitive *in-situ* hydrogels of rivastigmine-loaded lipid-based nanosystems for nose-to-brain delivery: Characterisation, biocompatibility, and drug deposition studies. International Journal of Pharmaceutics 2022; 620: 121720.

**How to cite this article:**
Nilwani KS, Wadkar HS and Narwade SP: Formulation and development of rimegepant loaded *in-situ* nasal gel for treatment of migrane by extended-release drug delivery. Int J Pharm Sci & Res 2025; 16(11): 2954-68. doi: 10.13040/IJPSR.0975-8232.16(11).2954-68.